Received: 13 October 2020 | Revised: 29 December 2020 | Accepted: 2 January 2021

DOI: 10.1111/tme.12761

ORIGINAL ARTICLE

TRANSFUSION MEDICINE WILEY

Detection and analysis of blood donors seropositive for syphilis

Adriana Attie[1] | Cesar de Almeida-Neto[1,2] | Steven S. Witkin[3,4] | Juliana Derriga[1] | Anna S. Nishiya[1,5] | Jerenice E. Ferreira[3,6] | Natalia de Souza Xavier Costa[7] | Nanci Alves Salles[1] | Tila Facincani[1] | Jose E. Levi[3] | Ester C. Sabino[2] | Vanderson Rocha[1,2,5] | Alfredo Mendrone-Jr[1,5] | Suzete C. Ferreira[1,5]

[1]Divisão de Pesquisa & Medicina Transfusional, Fundação Pró-Sangue Hemocentro de São Paulo, São Paulo, Brazil

[2]Disciplina de Ciências Médicas, Faculdade de Medicina da Universidade de São Paulo, São Paulo, Brazil

[3]Laboratorio de Investigação Medica em Virologia (LIM 52), Instituto de Medicina Tropical da Universidade de São Paulo, São Paulo, Brazil

[4]Department of Obstetrics and Gynecology, Weill Cornell Medicine, New York, New York

[5]Laboratory of Medical Investigation in Pathogenesis and Targeted Therapy in Onco-Immuno-Hematology (LIM-31), Department of Hematology, Hospital das Clínicas HCFMUSP, Faculdade de Medicina, Universidade de Sao Paulo, São Paulo, Brazil

[6]Centro de Patologia, Instituto Adolf Lutz, São Paulo, Brazil

[7]Departamento de Patologia, Faculdade de Medicina da Universidade de São Paulo, São Paulo, Brazil

Correspondence
Suzete C. Ferreira, Fundação Pró-Sangue Hemocentro de SP, São Paulo, Brazil.
Email: lom.suzy@gmail.com

Abstract

Background: The increasing incidence of syphilis worldwide has called attention to the risk of transmission by transfusion.

Aims: To determine the prevalence of active syphilis in blood donors and characterise the serological profile of syphilis-positive donors.

Methods: Samples positive for *Treponema pallidum* using the chemiluminescent microparticle immunoassay (CMIA) during blood donor screening from 2017 to 2018 were tested by the Venereal Disease Research Laboratory (VDRL) non-treponemal test and for anti-*T. pallidum* IgM by ELISA (Immunoassay Enzyme test for detection of IgM antibodies). The INNO-LIA Syphilis test (Line Immuno Assay solid test for confirmation antibodies to *Treponema pallidum*) was performed as a confirmatory test on samples that were positive on ELISA-IgM but negative on VDRL. ELISA-IgM (+) samples were also tested for *T. pallidum* DNA in sera by real-time polymerase chain reaction (PCR).

Results: Of 248 542 samples screened, 1679 (0.67%) were positive for syphilis by CMIA. Further analysis was performed on 1144 (68.1%) of these samples. Of those tested, 16% were ELISA IgM(+)/VDRL(+), 16.5% were ELISA IgM(–)/VDRL(+), 4.1% were ELISA IgM(+)/VDRL(–), and 63.4% were ELISA IgM (–)/VDRL(–). The INNO-LIA Syphilis test results were 33 (3%) positive, 2 (0.2%) undetermined and 12 (1%) negative. Of the 230 EIA-IgM(+) samples (20.1%), 5 (2.2%) were PCR positive. The prevalence of active syphilis in 2017 and 2018 was 0.1% and 0.07%, respectively, and overall prevalence of serologic markers for syphilis was highest among male, unmarried, 25–34-year-olds with a high school education and who were first-time donors.

Conclusion: There is a risk of transfusion-transmitted syphilis in blood banks that exclusively use the VDRL test for donor screening, as is currently the situation in some Brazilian blood centres, as well as in other blood centres around the world.

KEYWORDS
blood donors, IgM antibody, syphilis, *Treponema pallidum*, VDRL

1 | INTRODUCTION

Transmission of syphilis by blood transfusion has re-emerged in many countries as a threat to public health, especially among vulnerable populations. This likelihood requires a re-evaluation of current diagnostic tools and implementation of enhanced haemovigilance programmes.[1-10]

Since the mid-1980s, the need to continue serological screening for syphilis in blood donors has been debated. Although the American Association of Blood Banks has not required testing for syphilis since 1985, the US Food and Drug Administration (FDA) has not supported this change, and screening for syphilis has remained mandatory. The US Food and Drug Administration's (FDA's) position was to maintain syphilis testing as an indirect test of risk-related behaviour for susceptibility to human immunodeficency virus (HIV) infection rather than for the prevention of transfusion-transmitted syphilis.[11]

Brazil began serological screening for syphilis in blood donors using the VDRL test in 1969, and this test is still widely used.[1,9] Between 2010 and 2016, approximately 230 000 new cases of syphilis were reported, most of them located in the southeast region. According to data from the 2016 Epidemiological Bulletin, between 2014 and 2015, acquired syphilis increased by 32.7%, syphilis in pregnant women by 20.9% and congenital syphilis by 19%. In 2015, the total number of reported cases of syphilis in Brazil was 65 878. In the same period, the detection rate was 42.7 cases per 100 000 inhabitants, mostly among men (136 835, 60.1%). Although the increase in syphilis cases over time was evident, the Ministry of Health only announced that the country faced a syphilis epidemic in 2016.[4]

Transmission of *Treponema pallidum*, the causative agent of syphilis, through blood transfusion, although rare, is possible and is recognised as the third form of syphilis acquisition. Syphilis was the first transfusion-transmitted infection to be systematically investigated in blood donors following the implementation of serological screening in 1938.[5] Prior to the initiation of testing, more than 100 cases of transfusion-related syphilis were reported. After the start of serological screening in blood banks, there was a drastic reduction in the number of cases of transfusion-transmitted syphilis. In the last 40 years, only three cases of transfusion-related syphilis transmission have been reported.[6-8]

Despite this very low incidence, serological screening for syphilis remains mandatory in many countries, including Brazil. The significant reduction in transmission by transfusion was not only due to the introduction of syphilis screening in blood banks and improvement in donor recruitment but also because of its low incidence among blood donors in the 1990s and early 2000s, as well as *T. pallidum*'s inability to survive in refrigerated blood products.[9]

Transfusion transmission became so rare in developed countries in the late 1990s that the need for maintaining mandatory serological screening for syphilis in blood banks began to be questioned. However, cases of transfusion-transmitted syphilis may increase again because of the current resurgence of this infection, relaxation in donor selection criteria due to social pressure and non-compliance in donor screening interview responses. Platelet concentrates, frequently used in the treatment of patients with haemo-oncogenic disorders, although typically stored in pouches with oxygen in which *T. pallidum* cannot survive, may also be stored at ambient temperature (20–24°C) where the organism can remain viable. Thus, further discussion about whether or not mandatory donor screening should be enforced is worthwhile.[3,9]

Regarding recent efforts in transfusion medicine to improve safety conditions for donors, current screening protocols have limitations in the clinical interpretation of serological patterns, especially in asymptomatic blood donors.[1] The present study sought to determine the prevalence of active syphilis in blood donors, evaluate the reliability of the prevalent VDRL test and characterise individuals who were positive for *T. pallidum*.

2 | METHODS

2.1 | Study design

We conducted a retrospective cross-sectional analysis in 2019–2020 of samples from blood donations obtained between January 2017 and December 2018 at Fundação Pró-Sangue (FPS), the blood centre of São Paulo, Brazil, that were seropositive for syphilis. The study was approved by the Ethical Committee of Hospital das Clínicas at the University of São Paulo and the ethical review board of FPS (assent n°. 2.470.318) and was financially supported by FAPESP (2017/23028-9).

2.2 | Laboratory analyses, risk behaviours, donation type and motivational factors

All qualified candidate blood donors are routinely serologically screened for HIV types 1 and 2; human T-cell lymphotropic virus (HTLV) 1 and 2; hepatitis B (hepatitis B surface antigen [HBsAg] and total antibody to hepatitis B core antigen [anti-HBc]) and hepatitis C; syphilis and Chagas disease; and nucleic acid test (NAT) HIV, NAT-HBV (hepatitis B virus) and NAT-HCV (hepatitis C virus). All positive samples are stored in a repository and are available for subsequent in-depth analysis (retrovigilance). Donor samples that were positive for anti-treponema by a chemiluminescent microparticle immunoassay (CMIA) (Abbott Architect) during their initial screening were obtained and tested for IgM antibody to *T. pallidum* by ELISA (Euroimmun) and by the non-treponema-specific VDRL test (ANTIGEN-Omega Diagnostics). All samples positive for EIA-IgM or VDRL were tested by real-time PCR for *T. pallidum* DNA. The INNO-LIA Syphilis-Fujirebio Immunoblot test was also performed on samples that were EIA-IgM positive and VDRL negative (Figure 1).

As a routine in our blood centre, all donors who tested positive in any serologic screening test are recalled to collect a new sample to confirm the original results. They are also subjected to an interview about risk factors for syphilis and other transfusion-transmitted

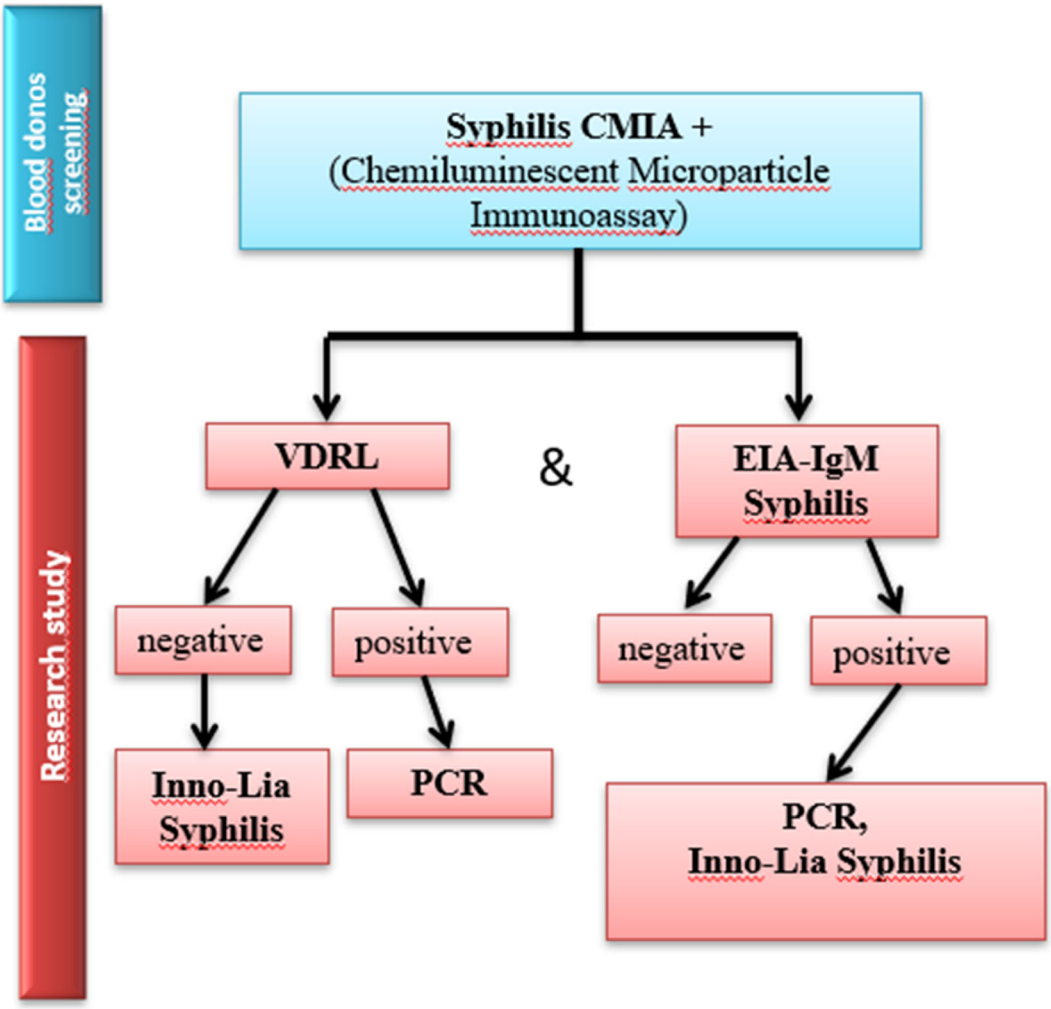

FIGURE 1 Flow chart of the study design. The CMIA test was used in the initial screening of blood donors. Positive samples were tested for VDRL, IgM antibody to *Treponema pallidum* (EIA-IgM), *T. pallidum* DNA by PCR and by the INNO-LIA Syphilis assay [Color figure can be viewed at wileyonlinelibrary.com]

diseases and their motivation to donate blood. The interviews are face to face, in a private room, using a standardised questionnaire and are conducted by trained physicians according to the Standard Operation Procedures of our institution. Donors who tested positive in the CMIA test were requested to provide repeat samples to confirm the results. When they returned for assay results, notification and counselling, the donors filled out a questionnaire to assess their risk factors for becoming infected with syphilis and motivations for blood donation. The questions asked included: "In the past 12 months, with how many different people have you had sex?" "Concerning your steady sexual partners, what was the frequency of condom use when you had sex?" and "Have you ever exchanged (given or received) money or drugs to have sex with someone?" Motivations for blood donation were classified as direct appeal, altruism and self-interest according to a previous publication.[12]

Donation type was classified as (i) first-time donation (a donation from an individual who had never donated in our blood centre), (ii) repeat donation (a donation from a person who donated at least twice in the last 12 months) and (iii) sporadic donation (a donation from someone who donated at least twice within an interval greater than 12 months).[13]

To detect *T. pallidum* DNA, 500 µl of serum were extracted by MagNa Pure Compact Nucleic Acid Isolation—Large Volume kit (Roche, Germany) in an automatized system MagNa Pure Compact (Roche, Germany), according to the manufacturer's protocol, and were subjected to real-time PCR. TaqMan was performed in StepOne Plus TM Real-Time PCR Systems (Life Technologies, Foster City, CA, USA). The primers and probes were designed using the assay of the design programme (Applied BioSystems, Carlsbad, CA, USA) targeting the polA gene of *T. pallidum*.[10]

2.3 | Statistical analyses

We used the SPSS 17 software (SPSS Inc/IBM Chicago, USA) for the statistical analyses. Sociodemographic variables included gender; age group; marital status; educational attainment; and first-time, repeat or sporadic donor status. Comparisons between the frequencies of the

TABLE 1 Demographic characteristics of CMIA-positive and syphilis IgM-positive blood donors

	CMIA+							*EIA-IgM* +						
	2017 (*n* = 617)		2018 (*n* = 527)		Total (*n* = 1144)		*p*-Value	2017 (*n* = 138)		2018 (*n* = 92)		Total (*n* = 230)		*p*-Value
Gender							0.6606							0.915
Male	337	(54.6%)	281	(53.3%)	618	(54.0%)		70	(50.7%)	46	(50.0%)	116	(50.4%)	
Female	280	(45.4%)	246	(46.7%)	526	(46.0%)		68	(49.3%)	46	(50.0%)	114	(49.6%)	
Age (years)							0.196							0.036
17–24	117	(19%)	110	(20.9%)	227	(19.8%)		34	(24.6%)	37	(40.2%)	71	(30.9%)	
25–34	188	(30.5%)	160	(30.4%)	348	(30.4%)		57	(41.3%)	35	(38%)	92	(40%)	
35–44	136	(22%)	88	(16.7%)	224	(19.6%)		32	(23.2%)	10	(10.9%)	42	(18.3%)	
45–54	102	(16.5%)	94	(17.8%)	196	(17.1%)		12	(8.7%)	6	(6.5%)	18	(7.8%)	
≥55	74	(12%)	75	(14.2%)	149	(13%)		3	(2.2%)	4	(4.3%)	7	(3%)	
Educational level							0.2273							0.5796
<Elementary school	43	(6.9%)	39	(7.4%)	82	(7.2%)		3	(2.3%)	3	(3.3%)	6	(2.6%)	
Elementary school	66	(10.7%)	73	(13.9%)	139	(12.2%)		12	(8.9%)	8	(8.8%)	20	(8.7%)	
High School	373	(60.5%)	307	(58.5%)	680	(59.5%)		96	(69.0%)	66	(71.5%)	162	(70.4%)	
College and above	135	(21.9%)	106	(20.2%)	241	(21.1%)		27	(19.8%)	15	(16.4%)	42	(18.3%)	
Marital status							0.02853							0.2795
Single	343	(55.6%)	273	(51.8%)	616	(53.8%)		92	(66.7%)	71	(77.2%)	163	(70.9%)	
Married	209	(33.9%)	173	(32.8%)	382	(33.4%)		35	(25.4%)	14	(15.2%)	49	(21.3%)	
Divorced/Separated	21	(3.4%)	28	(5.3%)	49	(4.3%)		3	(2.1%)	3	(3.3%)	6	(2.6%)	
Other	44	(7.1%)	53	(10.1%)	97	(8.5%)		8	(5.8%)	4	(4.3%)	12	(5.2%)	
Donation type							0.8913							0.6981
First time	604	(97.9%)	517	(98.1%)	1121	(98.0%)		135	(97.8%)	91	(98.9%)	226	(98.3%)	
Repeat	5	(0.8%)	4	(0.8%)	9	(0.8%)		2	(1.5%)	1	(1.1%)	3	(1.3%)	
Sporadic	8	(1.3%)	6	(1.1%)	14	(1.2%)		1	(0.7%)	0	(0.0%)	1	(0.4%)	
Donors who returned for notification and counselling	217	(35.2%)	190	(36.1%)	407	(35.6%)		39	(28.3%)	26	(28.3%)	65	(28.3%)	

TABLE 2 Motivations to donate blood among syphilis-positive blood donors

	Direct appeal		Altruism		Self-interest		
	n	(%)	*n*	(%)	*n*	(%)	*p*-Value
CMIA +	238	(58.5%)	157	(38.6%)	12	(2.9%)	
VDRL	67	(16.5%)	38	(9.3%)	3	(0.7%)	0.68
ELISA IgM +	36	(8.8%)	28	(6.9%)	1	(0.2%)	0.590
INNO-LIA +	73	(17.9%)	41	(10.1%)	3	(0.7%)	0.510
Active syphilis	36	(8.8%)	25	(6.1%)	0	0	0.328

sociodemographic characteristics and the treponemal/non-treponemal assay results were performed using the Pearson Chi-square (χ^2) test. Results were considered statistically significant at $p < 0.05$.

3 | RESULTS

Among 248 542 (123 851 in 2017 and 123 691 in 2018) samples screened, 1679 (0.67%) were positive in the CMIA assay. Of the 1144 (68.1%) positive patients available for inclusion in the study, 16.0% were EIA-IgM(+)/VDRL(+), 16.5% were EIA-IgM(–)/VDRL(+), 4.1% were EIA-IgM(+)/VDRL(–), and 63.4% were EIA-IgM(–)/VDRL(–). The INNO-LIA Syphilis test, performed as a confirmatory test in 47 (4.1%) samples that were EIA-IgM positive and VDRL negative, yielded 33 (3.0%) positive results, 2 (0.2%) that were inconclusive and 12 (1.0%) that were negative. Of the 230 EIA-IgM (+) samples, 5 (2.2%) were positive for *Treponema* DNA by real-time PCR.

In 2017, the prevalence of collected blood that was positive for syphilis screening tests was 0.77%. It was higher among men (54.6%) who were unmarried (55.6%) and between 25 and 34 years old (30.5%) with a high school education (60.5%) and who were first-time donors (97.9%). In 2018, the prevalence of blood units positive for syphilis screening tests was 0.62%. Among the EIA-IgM-positive samples, 10.4% were positive for antibody to HBV anti-Core, 1.1% for antibody to HIV, 1.5% for anti-HTLV-1/2 and 1.1% for NAT-HIV (Table 1).

Among the 1144 donors who returned following notification for counselling, 407 (35.6%) completed the questionnaire, and 33 (2.9%) responded affirmatively to the question, "Have you ever exchanged (given or received) money or drugs to have sex with someone?" Of these, 57.7% were above 45 years of age, 53.3% were married, 53.3% graduated from high school, and 100% were first-time donors.

In Table 2, we show that, among motivation choices, a direct appeal was the most frequent response (58.5%), followed by altruism (38.6%).

Associations with condom usage are shown in Table 3. About a third of female and a quarter of male donors never used condoms during sex. The prevalence rate for syphilis was 55.5% among donors who did not use condoms, 25.3% for those who used condoms sometimes and 16% for those who always used condoms ($p < 0.044$). Among the women positive for syphilis, 15.5% were between 45 and 54 years old, 28.2% were married, 30.2% had a high school education level, and 56.6% were first-time donors ($p < 0.0001$) (Tables 3 and 4).

4 | DISCUSSION

In blood donations provided to Fundação Pró-Sangue Hemocentro de São Paulo, one of the largest blood banks in the city of São Paulo, between 2015 and 2017, there was an apparent increase of 24% in the detection of syphilis-associated markers, from 0.62% in 2015 to 0.73% in 2016 and 0.77% in 2017, followed by a small decline to 0.62% in 2018 ($p < 0.0001$). A similar increase was reported in 2014–2015 in the United States (3). Also paralleling our findings, the donors in their study with the highest rate of a positive test were men, unmarried, between 25 and 34 years old, with a high school education and were first-time blood donors (3).

The prevalence of active syphilis among blood donors seen at our centre between 2017 and 2018 was 0.09%. We identified 35 cases that were negative for syphilis by VDRL but were anti-*T. pallidum* IgM and INNO-LIA positive. These findings are consistent with a study by Moore et al, who showed that non-treponemal tests for primary syphilis infection were negative in 30%–50% of infected individuals.[12-17] They strongly suggest that there remains a risk for transfusion-transmitted syphilis in those blood banks that exclusively use the VDRL test for syphilis donor screening. In addition, the detection of co-infection with HIV, HBV or HTLV-1/2 in 2.2%, 10.4% and 1.5% of syphilis-positive cases, respectively, suggests that the application of treponemal-specific tests are also relevant for the prevention of the transfusion-related transmission of other sexually transmitted infections.[18]

Our demonstration that 2.2% of EIA-IgM-positive donors had *T. pallidum* DNA in their circulation is consistent with a previous investigation by Dow et al and suggests that this organism might still be present in some individuals despite evidence of an antibody response. It must be acknowledged that detection of *T. pallidum* DNA cannot distinguish between the presence of viable or dead organisms.[5,18] Similar results were found in our previous study conducted in 2014, where we detected 2 (1.02%) cases positive for *T. pallidum* DNA from a total of 197 blood samples from donors positive for syphilis.[10] However, the routine use of a nucleic acid amplification test for syphilis is not recommended for all blood donors due to expense, the need for trained personnel and uncertainty about organism viability. In addition, treponema-specific antibody tests appear to be sufficient to identify infected individuals and prevent transfusion transmission.[5,10]

Among our blood donors who were positive for syphilis, it was not surprising that the highest risk of active syphilis infection occurred in those who did use condoms. This observation was also previously described by Hopkins et al in 2004.[15] The preferential screening for

TABLE 3 Associations with condom usage in blood donors

	How often did you use condoms (*n* = 407)						
	Never		Sometimes		Always		*p*-Value
Gender[a]							0.0555
Male	98	52.1%	51	27.1%	39	20.8%	
Female	128	62.1%	52	25.2%	26	12.6%	
Age (years)[a]							<0.0000001
17–24	20	28.2%	29	40.8%	22	31.0%	
25–34	53	48.2%	36	32.7%	21	19.1%	
35–44	44	60.3%	21	28.8%	8	11.0%	
45–54	61	73.5%	10	12.0%	12	14.5%	
≥55	48	84.2%	7	12.3%	2	3.5%	
Education[a]							0.0283
<Elementary school	30	81.1%	7	18.9%	0	0.0%	
Elementary school	24	60.0%	10	25.0%	6	15.0%	
High school	119	52.0%	67	29.3%	43	18.8%	
College and above	51	59.3%	19	22.1%	16	18.6%	
Marital status[a]							<0.0000001
Single	76	39.8%	66	34.6%	49	25.7%	
Married	111	74.5%	28	18.8%	10	6.7%	
Divorced/separated	15	88.2%	1	5.9%	1	55.9%	
Other	24	64.9%	8	21.6%	5	13.5%	
Donation type[a]							0.8443
First time	223	57.3%	101	26.0%	65	16.7%	
Repeat	1	50.0%	1	50.0%	0	0.0%	
Sporadic	2	66.7%	1	33.3%	0	0.0%	

[a]Total may be missing two values.

TABLE 4 The association between detection of syphilis, condom usage and specific behaviours

	CMIA +		ELISA IgM +		
	n	(%)	*n*	(%)	*p*-Value
How often did you use condoms when you had sex?					0.0593
Never	226	(57.4%)	27	(42.9%)	
Sometimes	103	(26.1%)	25	(39.7%)	
Always	65	(16.5%)	11	(17.5%)	
Have you ever exchanged money or drugs to have sex with someone?					0.145
Yes	33	(8.1%)	2	(3.1%)	
No	374	(91.9%)	63	(96.9%)	

sexually transmitted diseases in blood donors who engage in unprotected sexual intercourse is certainly warranted.

A direct appeal for blood was the most frequent motivation for blood donation, followed by altruism, similar to our previous findings.[19] Only 2.9% of donors were motivated by self-interest. More research is needed as to why individuals who are at elevated risk for syphilis and othersexually transmitted diseases (STDs) volunteer to donate blood. The availability of STD testing at the blood centre might be an additional motivating factor that overlaps with more socially accepted responses such as altruism and direct appeal.[19-21]

An advantage of our study was the ability to analyse findings from a large number of individuals, all of whom underwent a similar testing protocol from a single major specialised service. This increases the probability of uniform handling of all specimens. One limitation of our study is that the bacterial load for *T. pallidum* in donated blood is low because donors are typically healthy and asymptomatic individuals. Therefore, we

were unable to further analyse the PCR-positive samples for other treponemal genes. This would have been of value to provide evidence of the possible presence of intact organisms. This limitation was also reported by Ferreira et al in 2014.[12] Other limitations include the absence of data on the length of syphilis infection and mode of acquisition in positive donors.

Since 2016, serological evidence of syphilis has become the most prevalent marker for infectious disease found in blood donors at our institution. Continuous monitoring of the profile of syphilis-infected donors at this time of re-emergence of the infection is useful and relevant not only for blood banks but also as a reflection of the epidemiological status of syphilis in the community. Availability of these data can contribute to the refocusing of health policies and priorities. Our demonstration that 3-% of donors with acute phase syphilis antibodies were negative in the VDRL test strongly suggests that non-treponemal tests are not ideal for screening blood donors. In addition to a lack of sensitivity, results of these assays are subjective and require interpretation by an experienced technician.[10]

In conclusion, we emphasise that, due to the increased incidence of syphilis among blood donors worldwide, it is clearly necessary that new syphilis screening guidelines for blood donors be established to maximise transfusion safety.

ACKNOWLEDGMENTS

All authors fulfilled the following four criteria: substantial contributions to the conception or design of the work; and/or the acquisition, analysis, or interpretation of data for the work; and/or drafting the work or revising it critically for important intellectual content. All authors gave final approval of the version to be published and agreed to be accountable for all aspects of the work and in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. **Adriana Attie** performed the research, contributed essential reagents or tools, analysed data, wrote the manuscript and reviewed and edited text and tables. **Cesar de Almeida-Neto** designed the research study, performed the research, analysed data, wrote the manuscript and reviewed and edited text and tables. **Steven S. Witkin** analysed data, wrote the manuscript and reviewed and edited text and tables. **Juliana Derriga** contributed essential reagents or tools and analysed data. **Anna S. Nishiya** contributed essential reagents or tools and analysed data. **Jerenice Esdras Ferreira** analysed the data and reviewed and edited the text. **Natalia de Souza Xavier Costa** analysed the data and reviewed and edited the text. **Nanci Alves Salles** contributed essential reagents or tools. **Tila Facincani** reviewed and edited the text. **Jose E. Levi** reviewed and edited the text. **Ester C. Sabino** reviewed and edited the text. **Vanderson Rocha** reviewed and edited the text. **Alfredo Mendrone-Jr** analysed the data and reviewed and edited the text. **Suzete C. Ferreira** designed the research study, performed the research, analysed data, wrote the manuscript and reviewed and edited the text and tables.

CONFLICT OF INTEREST

The authors have no competing interests.

ORCID

Cesar de Almeida-Neto https://orcid.org/0000-0002-8490-4634
Jose E. Levi https://orcid.org/0000-0002-3557-2717

REFERENCES

1. Owusu-Ofori AK, Parry CM, Bates I. Transfusion-transmitted syphilis in teaching hospital, Ghana. *Emerg Infect Dis*. 2011;17:2080-2082.
2. Waugh M. The centenary of *Treponema pallidum*: on the discovery of Spirochaeta pallida. *Skinmed*. 2005;4:313-315.
3. Avelleira JCR, Bottino G. Syphilis: diagnosis, treatment and control. *An Bras Dermatol*. 2006;81:111-126.
4. Goh BT. Syphilis in adults. *Sex Transm Infect*. 2005;81:448-452.
5. Dow BC, Franklin IM, Munro H, Gunson R. Syphilis nucleic acid testing: usefulness in syphilis confirmation? *Transfusion*. 2010;50:737-739.
6. Ferreira C, Ferreira W, Motta C, Vasquez F, Pinto A. Reatividade do teste VDRL em bolsas de sangue da Fundação de Hematologia e Hemoterapia do Amazonas-HEMOAM, os custos decorrentes do descarte e a estimativa de prevalência de sífilis em doadores de sangue do estado do amazonas. *DST J Bras Doenças Sex Transm*. 2006;18:14-17.
7. Chambers RW, Foley HT, Schimidt PJ. Transmission of syphilis by fresh blood components. *Transfusion*. 1969;9:32-34.
8. Perkins HA, Busch MP. Transfusion associated infections: 50 years of relentless challenges and remarkable progress. *Transfusion*. 2010;50:2080-2099.
9. Oliveira VM, Verdasca IC, Monteiro MC. Syphilis detection using ELISA and VDRL tests on blood donors at the blood center of Guarapuava, State of Paraná. *Rev Soc Bras Med Trop*. 2008;41:428-430.
10. Ferreira SC, Almeida-Neto C, Nishiya AS, et al. Prevalence of *Treponema pallidum* DNA among blood donors with two different serologic tests profiles for syphilis in Sao Paulo, Brazil. *Vox Sang*. 2014;106:376-378.
11. WHO. Global prevalence and incidence of selected curable sexually transmitted infections: overview and estimates. *World Health Organization Department of HIV/AIDS, 2001*. Geneva, Switzerland: World Health Organization; 2001.
12. Ferreira SC, Almeida-Neto C, Nishiya AS, et al. Demographic, risk factors and motivations among blood donors with reactive serologic tests for syphilis in São Paulo, Brazil. *Transfus Med*. 2014;24:169-175.
13. Agência Nacional de Vigilância Sanitária (Brasil). 5° Boletim Anual de Avaliação Sanitária em Serviços de Hemoterapia. Novembro 2013. Disponível em:. Acesso em: 31 maio. 2014.
14. Moore M, Knox J. Sensitivity and specificity in syphilis serology: clinical implications. *South Med J*. 1965;58:963-968.
15. Hopkins S, Lyons F, Coleman C, Courtney G, Bergin C, Mulcahy F. Resurgence in infectious syphilis in Ireland: an epidemiological study. *Sex Transm Dis*. 2004;31(5):317-321.
16. Shrama S, Sethi S, Garg S, Lamba DS, Sharma RR, Marwaha N. Comparative evaluation of enzyme-linked immunosorbent assay with rapid plasma reagin for screening of syphilis in blood donors. *Asian J Transfus Sci*. 2018;12:165-168.
17. Orton S. Syphilis and blood donors: what we know, what we do not know, and what we need to know. *Transfus Med Rev*. 2001;15:282-292.
18. Salles NA, Sabino EC, Barreto CC, Barreto AM, Otani MM, Chamone DF. The discarding of blood units and the prevalence of infectious diseases in donors at the Pro-Blood Foundation/Blood Center of São Paulo, São Paulo, Brazil. *Rev Panam Salud Publica*. 2003;13:111-116.
19. Carneiro-Proietti AB, Sabino EC, Sampaio D, et al. Demographic profile of blood donors at three major Brazilian blood centers: results from the international REDS-II study, 2007 to 2008. *Transfusion*. 2010;50:918-925.

20. Cavalcante EG, Araujo MA, Galvão MT, de Moura HJ, Gondim AP, da Silva RM. Sexually transmitted infections associated syndromes assisted in the primary health care in Northeast, Brazil. *BMC Public Health*. 2012;12:595.
21. Gonçalez TT, Di Lorenzo Oliveira C, Carneiro-Proietti ABF, et al. Motivation and social capital among prospective blood donors in three large blood centers in Brazil. *Transfusion*. 2013;53(6):1291–1301. http://dx.doi.org/10.1111/j.1537-2995.2012.03887.x.

How to cite this article: Attie A, de Almeida-Neto C, S. Witkin S, et al. Detection and analysis of blood donors seropositive for syphilis. *Transfusion Medicine*. 2021;1–8. https://doi.org/10.1111/tme.12761